ART. XIV. LARVAL DEVELOPMENT OF THE EASTERN NARROW-MOUTHED FROG, MICROHYLA CAROLINENSIS (HOLBROOK), IN LOUISIANA

# By Grace L. Orton

# (Plate I)

While engaged in herpetological collecting in the vicinity of Baton Rouge, Louisiana, from May 30 to June 20, 1945, I obtained several series of tadpoles of *Microhyla carolinensis* (Holbrook). These specimens, ranging from early developmental stages to metamorphosing individuals, provide information on a number of features of the larval morphology and development of this frog, and the publication of a separate report on them appears to be warranted.

My work in Louisiana was greatly aided by the privilege of using laboratory facilities in the Department of Zoology of Louisiana State University. For this, and for other kindnesses, I am especially indebted to Prof. William H. Gates, Prof. Ellinor H. Behre, and Dr. Ira D. George.

The first accounts of the life history of Microhyla carolinensis appear to be those of Ryder (1891) and Deckert (1914). Both were concerned particularly with the developmental rate, and included little morphological data. The tadpole was first described in detail by Wright (1929). His account was illustrated by a figure of the mouthparts and a small photograph of the tadpole. More descriptive details were included in his later work (1932), with the same figure of the mouthparts and two dorsal-view photographs of the tadpole. His work on this species was based largely on material from the Okefinokee Swamp. Noble (1929) provided a few details on embryonic development, with figures of young embryos and a ventral view of an advanced tadpole. Wright and Wright (1933, 1942) gave a very brief description of the tadpole, and repeated the earlier (1929) illustrations, with one additional lateral-view photograph. Louisiana tadpoles of this frog have not been described previously, the ventral color pattern has not been adequately analyzed and illustrated, nor have the details of spiracle formation been reported. These and other points are treated in the present paper.

The Louisiana specimens upon which this study is based are as follows:

From in or immediate vicinity of Baton Rouge, East Baton Rouge Parish: CM 24213 (55), collected June 5; CM 24214, developmental series from eggs collected June 5; CM 24219 (163), collected June 6;

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CM 24267 (4), collected June 11; CM 24313 (1), collected June 16; CM 24358 (4), collected June 19.

From one mile northeast of St. Francisville, West Feliciana Parish: CM 24276 (1), collected June 12.

A series of six early developmental stages (CM 24214) was obtained from eggs collected on the morning of June 5, in shallow water in a marshy area along the lake adjacent to the State Capitol grounds. The eggs were still in a pre-gastrula stage of development when collected, and were presumed to have been laid during the previous night. An unusually large chorus of *Microhyla* had been heard there during the evening. The eggs were small, measuring  $\pm$  1.3 mm. in diameter (excluding the jelly), and were in small groups of about twenty to forty, each group in a single layer floating at the surface. The jelly of each egg was distinctly flattened dorsally, as noted by Wright (1932) and others. Although the eggs were sufficiently adhesive to attach to each other under normal conditions in the field (and to collecting instruments), the adhesion was not sufficiently strong to prevent the group from breaking up into individual eggs when handled.

# TABLE I

Early Larval Development of *Microhyla carolinensis* (based on CM 24214)

	Time of	
Unit	Preservation	Remarks
а	June 5, 12:30 р.м.	Eggs; pre-gastrula.
b	June 6, 9:30 A.M.	Hatching; total length $\pm$ 4 mm. Tail forming, but shorter than body; head structures still very in- complete, gill region swollen but gills not yet formed, paired adhesive organs well developed, mouthparts and eyes not yet externally visible.
С	June 6, 7:00 p.m.	Total length $\pm$ 5 mm. Tail well developed; body relatively shorter and thicker, head broader, eyes indicated, external gills well developed, adhesive organs widely separated, mouth better defined but labial structures not yet evident; opercular folds not yet distinct.
d	June 7, 9:00 а.м.	Total length $\pm$ 6 mm. Body assuming typical mi- crohylid shape; eyes well developed; opercular folds completed, gills covered; spiracle well developed, in advanced specimens opening about halfway be- tween posterior edge of gill chamber and anus; mouth subtriangular, some trace of lip formation.

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е	June 7, 6:30 р.м.	Total length $\pm$ 6.5 mm. Body of typical microhylid			
		shape; pigmentation better developed, individual			
		melanophores clearly visible; spiracle opening			
		farther back, nearly in its final position; labial folds			
		quite well developed.			
f	June 8, 10:00 p.m.	Total length $\pm$ 7.5 mm. Spiracle opening below anus;			
		adhesive organs still evident as pigmented spots, but			
		no longer distinctly projecting.			

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General information on the developmental series is summarized in Table I. The principal gap in this series is between units c and d, where the critical early phases in operculum and spiracle formation were missed. In the least advanced specimens in unit d, the opercular folds from the two sides have already joined transversely to form a single short median fold. The narrowing of the opening to form the single median spiracle of this species, and its posteriad migration, can be traced in other specimens (Text fig. 1). Migration of the spiracle continues through elongation and



FIG. 1. Four stages in the formation of the spiracle and its posteriad migration in *Microhyla carolinensis*. CM 24214.

enlargement of the gill chamber and spiracular tube until, in tadpoles of about 7.5 mm. in total length, the opening has reached its final position, below and posterior to the anus (Pl. I, fig. 1). By the time this size is attained, the adhesive organs have flattened out, but they persist as pigmented spots until a total length of about 9 mm. is reached.

The developmental rate of the eggs of *Microhyla carolinensis* is probably influenced by temperature to some extent. Ryder (1891) and Deckert (1914), in North Carolina and Florida, respectively, found that the eggs of this frog hatch about three days after they are laid. Wright (1932), working in the Okefinokee, estimated that hatching took place after three days on one occasion; eggs laid a few days later hatched in about a day Annals of the Carnegie Museum

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and one half. The Louisiana series reported upon in this paper hatched in approximately a day and one half.

The wide range in size of the tadpoles in some of the series collected in Louisiana confirms reports by previous authors that M. carolinensis has a prolonged breeding season. The 55 specimens in CM 24213 (taken on June 5, at the same time and place eggs were found) range from 5.5 to 24 mm. in total length, with no sharply defined size groups. It should be pointed out, however, that this particular series is not a random sample of the population, for, after initial sampling, effort was directed toward obtaining the larger tadpoles. Another series, CM 24219 (collected on June 6, in a ditch southwest of the Louisiana State University campus), contains two well defined size groups: 90 larvae, which obviously represent more than one period of breeding, range from 5 to 12.5 mm. in total length (head and body,  $\pm 2-6$ ); and 73 larvae, including two metamorphosing specimens, vary from 27.5 to 34.5 mm. (head and body, 10-12). On June 11, two larvae of 21 and 38 mm., total length, and two transforming specimens were taken; on June 16, a recently-hatched larva of 6 mm. total length was found; and on June 19, larvae of 6.5 to 10.5 mm. in total length were obtained.

By combining the data from the several series of tadpoles, some information on ontogenetic changes becomes apparent and can be summarized. In the material at hand, the hind leg buds become evident as minute, rounded bumps when the tadpole attains a total length of about 8 to 10 mm. These structures increase very slightly in distinctness up to a total length of about 15 to 16 mm., at which time they begin to elongate slightly. In tadpoles of about 19 to 20 mm., the leg buds have enlarged to about 1 mm. in length. At about 21 to 24 mm., the first traces of joint and foot differentiation can be detected, and the leg bud length is about 1.5 mm. Above a total length of 24 mm., leg size increases abruptly, although there is still a general correlation between total length and leg size. In the specimens examined, those under 30 mm. in total length have leg buds of 2.5 to 6.5 mm. in length, and those over 30 mm. have leg buds of from about 5 mm. up to the transformation size of  $\pm$  10 mm. Since the snout-vent length of the tadpole at transformation is variable (Wright, 1932), an exact correlation of total length and leg length is not to be expected. It is significant to note that between the time the leg buds reach the readily measurable length of one millimeter and the time of metamorphosis, the total length of the tadpole increases as much as 60 per cent, perhaps much more in some individuals. Some increase in total length apparently continues up to very near metamorphosis.

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The development of the striking larval color pattern of M. carolinensis can also be traced. The coloration is essentially uniform blackish until a total length of 9 to 11 mm. is reached, when some traces of light ventral spotting become evident in some specimens. The first elements of the pattern to become distinct—and the most constant in occurrence and outline-are two pairs of light spots on the sides, which enlarge and become oblique longitudinal or slightly curved streaks. These lateral spots are usually well defined in 16 mm. larvae, but the ventral pigmentation shows much variation in distinctness. A well defined pattern of ventral spotting is present in most of the larvae over 24 mm. in total length. In 73 larvae having a total length of 27.5 to 34.5 mm. (CM 24219), the skin over the abdomen is opaque and its ground color varies from very dark (blackish in life) to pale bluish-gray, depending upon the state of contraction of the melanophores and probably also upon differences in the actual amount of dark pigment present in individual larvae. Most of the specimens are intermediate between the two color extremes. The bellies of all but about a half-dozen of the tadpoles are quite heavily spotted, and most of the exceptional specimens would be too pale to show pattern elements distinctly. The basic pattern of light spots appears to be as follows: two pairs of oblique longitudinal streaks along the sides; a pair of transverse semicircular spots across the belly; anterior to these a single longitudinal median streak, forked posteriorly; and a pair of short streaks along the base of the spiracular tube, near its opening. In about 15 of the 73 larvae the pattern is very diagrammatic, with all the basic elements sharply defined and not obscured by additional spotting (Pl. I, fig. 3). In the more heavily marked majority, some part of the basic pattern is still, however, usually recognizable (Pl. I, fig. 4).

The development of the mid-lateral light stripe on the side of the tail appears to begin about the same time the ventral spot pattern begins to differentiate. This light stripe is present and conspicuous in all large (over 20 mm. in total length) larvae in the present collection.

Since the differentiation of the color pattern and increase in size continue well after the developing leg buds attain an easily measurable length, it seems apparent that, in this species at least, the term "maturity" as conventionally applied to tadpoles with evident leg buds is a misnomer.

The Carnegie Museum collection includes four tadpoles of *Microhyla* carolinensis from Virginia (CM 19053 and 23784), and two from Florida (CM 8705). They are all of similar size, ranging from 21 to 23.5 mm. in total length (head and body, 7.5-9), with hind leg buds of  $\pm 1$  mm. in

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length. With respect to extent of leg development, they closely parallel specimens of comparable total length from Louisiana. The ventral color pattern is evident in all, and is especially conspicuous in the two Florida specimens.

To facilitate comparison with specimens from other parts of the range, a detailed description of larval *Microhyla carolinensis* in Louisiana is provided. It is based largely on the series of 73 large larvae included under CM 24219.

*Description:* (Pl. I, fig. 1). Head and body broad, flattened dorsally, belly moderately deep but not globose. Eyes small, on sides of the broad flat head. External nares not apparent until near metamorphosis, when they open very close together on top of snout. Skin thick and opaque on body and basal third or more of tail. Anus median. Spiracle median, far back, its tubular opening below and slightly posterior to anus. Tail length somewhat variable, usually about 1.5 times head and body length; tail tapers gradually, tip pointed, fins moderately developed, slightly deeper than musculature at point halfway back on tail.

*Mouthparts:* (Pl. I, fig. 2). Mouth opening small, terminal; upper jaw with a pair of broad rounded lateral flaps overhanging lower jaw and separated medially by a deep concavity; lower jaw with a pair of transverse basal segments joined by a soft outwardly-directed, median loop-like element; a short labial fold along basal segment of jaw, extending from base of median segment back to join lateral base of upper jaw flap; jaw margins soft, no horny beaks, a few specimens with traces of papilla-like processes on margins of upper jaw flaps.

*Color* (*in life*): Body dark blackish, dorsal surface very finely stippled with yellow, with no evident pattern; sharp line of demarkation from eye to tip of snout between yellowish above and black of side of head. Upper jaw flaps usually edged with black. Sides darker than dorsum and with less yellow stippling. Skin of belly opaque, its ground color blackish to pale blue-gray; conspicuous lateral and ventral spots very pale orange, darkedged. Tail musculature black, dorsal edge stippled with yellowish on basal third of tail; a sharply-defined mid-lateral light stripe very prominent on basal third of tail where skin is thick and opaque, much less distinct distally. Tail fins heavily pigmented with black along the half bordering the musculature; marginal half of both upper and lower fins usually unpigmented.

The distinctive pattern and the pale orange color of the ventral spots constitute good field characters for the identification of this tadpole.

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The largest specimen in the Okefinokee material reported upon by Wright (1932) had a total length of 26.4 mm. (head and body, 10 mm.). In the Louisiana collection there are 74 tadpoles which exceed this size. They comprise 73 specimens in CM 24219, with total lengths of 27.5 to 34.5 mm. (head and body, 10-12), and one tadpole in CM 24267, with a total length of 38 mm. (head and body, 13.5). The body lengths of transforming specimens were given by Wright as 8.5 to 12 mm. Measurements of the four specimens in metamorphosis obtained at Baton Rouge are:

CM number	Date	Snout-vent length	Total lengt	h
24219	June 6	11.5	27.0	
24219	June 6	11.5	28.5	
24267	June 11	12.5	28.0	(tail beginning
24267	June 11	13.0	32.0	to shorten)

In all four of these specimens the front legs are out, and metamorphosis of the head structures has begun. In the absence of data from larger series and from more localities, it is not certain whether the size differences indicated by these measurements of tadpoles and transforming young have a valid geographic basis, or whether they may be correlated mainly with local or seasonal factors.

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#### EXPLANATION OF PLATE I

Louisiana tadpoles of Microhyla carolinensis (Holbrook).

- FIG. 1. Lateral view. Total length, 30 mm. CM 24219.
- FIG. 2. Mouthparts. Lateral flap of upper jaw raised on one side to show labial fold along lower jaw. CM 24219.

# FIGS. 3, 4. Variation in ventral color pattern. Fig. 3 shows basic pattern of ventral spots. Fig. 4 shows an extreme variant. CM 24219.